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Available in: http://www.redalyc.org/articulo.oa?id=289029240062

Acta Scientiae Veterinariae,
ISSN (Printed Version): 1678-0345
ActaSciVet@ufrgs.br
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www.redalyc.org
Non-Profit Academic Project, developed under the Open Acces Initiative
Clinical Effectiveness of Autohemotherapy as an Adjunct Treatment of Canine Parvovirus

Olivia Maria Moreira Borges¹, Almir Pereira de Souza², Rodrigo de Souza Mendes¹, Kamila Nunes de Araújo³, Leonardo Mendes Torres³ & Alinne Kattia Fernandes Pereira Dantas³

ABSTRACT

Background: Canine parvovirus is highlighted among the gastroenteric diseases that are common medical occurrences in small animals due to its clinical evolution. Although the treatment is often successful, the success rate has remained unchanged over the years, reflecting a clear need for searching more efficient protocols in order to reduce hospitalization, treatment cost and increase survival rates. Autohemotherapy efficacy has been demonstrated for several diseases and identified as a stimulating tool and immune mediator. Thus, the objective of this research was to determine the clinical effectiveness of autohemotherapy as an adjunct in the treatment of dogs suffering from parvovirus.

Materials, Methods & Results: Twenty dogs suffering from parvovirus, diagnosed via immunochromatography, were distributed (n = 10) into two groups: autohemotherapy (GAHT) and control (GCO). Both groups were treated with the appropriate support therapy for hemorrhagic gastroenteritis (GHE), while the dogs of the GAHT group were also treated with the adjunct autohemotherapy (AHT). The AHT consisted of the administration of blood collected by jugular venipuncture, without anticoagulant, immediately injected in equal proportion in the semitendinosus and semimembranosus muscle region. The blood volume injected was based on animal weight: up to 5 kg/2.5 mL; between 5-10 kg/5 mL; 10-15 kg/7.5 mL; above 15 kg/10 mL. The dogs of the control group were injected a NaCl 0.9% solution instead of blood. The vital parameters, heart rate (HR), respiratory rate (RR), and rectal temperature (RT), as well as the clinical parameters, mucous color, dehydration, level of consciousness, abdominal tenderness, anorexia, diarrhea and vomiting were evaluated and recorded. The hematological and parasitological stool analyses were also performed. The parameters were measured upon hospital admission; time zero (M0) and every 12 h during 48 h (M12, M24, M36 and M48). The vital parameters were submitted to ANOVA or Friedman test. For the hematological variables and remission of clinical signs of paired samples the t test or Wilcoxon test was performed. The comparison of averages/medians was performed by either the t test or Mann-Whitney test. The level of significance used was 5% (P < 0.05). All clinical parameters that indicated remission of the disease were statistically significantly different in the GAHT dogs, which lead to early hospital discharge. The GAHT animals exhibited more effective recovery signs, evidenced by the reduction of recovery time in half. The hospital discharge record shows that one dog was discharged after 33 h and another two dogs, after 46 h.

Discussion: During the hospitalization, complete remission of clinical signs initially cited was observed, but this was not homogeneous, as can be seen in GCO, in which three animals needed to be treated for a longer period. Two of them presented endotoxemia and one died 72 h and 70 h after starting the treatment. The comparison shows that animals of GAHT recovered more effectively, evidenced by the recovery time half as long as that needed for the dogs of the GCO. Thus, the autohemotherapy proved to be clinically effective, and resulted in premature recovery of patients suffering from parvovirus. Furthermore, no side effects or systemic organic impairment was observed in this treatment, revealing benefits as an alternative therapy and adjunct to treatment of this disease.

Keywords: autohemotherapy, diarrhea, parvovirus, dog, early recovery.
INTRODUCTION

Gastroenteric diseases are common medical occurrences in small animals, whose typical clinical signs are vomiting and diarrhea [6]. Since 1970, viral enteritis is considered one of the most common causes of infectious diarrhea in dogs, thus becoming one of the most common diseases of the species [14]. The parvovirus stands out among the juvenile canine viruses, since it presents severe gastroenteric symptoms, resulting in high rates of clinical complications and mortality [7].

Although the appropriate treatment is often successful, the success rate has remained unchanged over the years, reflecting a clear need to investigate more effective protocols in order to further reduce morbidity rate, hospitalization time and treatment cost, and to increase the survival rate, as well.

Autohemotherapy arises as one of several therapeutic protocols adjuvants to treat this disease. Although little studied, autohemotherapy has shown clinical efficacy in the treatment of several diseases [9,21,23,25], being pointed out as an immune mediator and stimulatory tool [24], assisting in the potentialization of the organic defenses against the debilitating infection. Thus, the aim of this study was to determine the clinical effectiveness of autohemotherapy as an adjuvant in the treatment of dogs suffering from parvovirus.

MATERIALS AND METHODS

The experimental site and the animals

The study was conducted in the Small Animal Clinical Sector of the Veterinary Hospital of the Federal University of Campina Grande, Campus de Patos-PB (HV/UFCG) and in the Veterinarian Medical Center Dr. Leonardo Torres (CMVLT), Patos-PB.

A total of twenty dogs seen at the outpatient care unit, regardless of age, sex or breed, with clinical signs of hemorrhagic gastroenteritis caused by Parvovirus, confirmed by immunochromatography kits were used.

Experimental Design

The animals were randomly divided into two groups (n = 10), the autohemotherapy (GAHT) group and the control (GCO) group. After confirming the involvement of parvovirus, both groups were treated with the appropriate therapy for hemorrhagic gastroenteritis (GHE), while the dogs of the GAHT group were also treated with the adjunct autohemotherapy (AHT).

The treatment which was adapted and consisted of Ringer’s Lactate Solution and/or 0.9% NaCl solution for correction of dehydration; Cimetidine hydrochloride 5 mg/kg/SC/BID as H2 antagonist; Citrate Maropitant 1 mg/kg/SC/SID, as centrally acting antiemetic; Enrofloxacin 2.5%, 5 mg/kg/SC/BID as antibiotics; Dipyrone 25 mg/kg/IV/SID as analgesic and antipyretic; and multivitamin, 20 mL/infusion [1].

Description of the AHT technique

The AHT consisted of immediately injecting the blood collected by jugular venipuncture with sterile syringes without anticoagulant, respecting a fractionation of equal amounts in the semitendinosus or semimembranosus muscles of the right and left limbs region, which were previously shaved and thoroughly cleaned. The volume administered was based on animal weight, determined in previous pilot studies: 2.5 mL for animals up to 5 kg; 5 mL/between 5 and 10 kg; 7.5 mL/10-15 kg; and 10 mL/above 15 kg. The dogs of the control group (GCO) underwent the same procedure, but the blood was replaced by NaCl 0.9%.

Evaluated clinical parameters

The vital parameters: heart rate (HR), respiratory rate (RR) and rectal temperature (RT), as well as the clinical; color of mucous membranes, dehydration, level of consciousness, abdominal tenderness, anorexia, diarrhea, and vomiting were evaluated and recorded [19]. Parameters were measured at the time of outpatient care (M0) and every 12 h, for a total of two evaluation days (M12, M24, M36 and M48). The patient was discharged after remission of the gastrointestinal symptoms.

Statistical Analysis

Data normality was checked using the Shapiro-Wilk test. The clinical data obtained were subjected to analysis of variance (ANOVA) or Friedman test. Wilcoxon test or t-test was used for remission of clinical signs of paired samples. The comparison of averages/medians between CGO and GAHT was performed by the t-test or Mann-Whitney test as indicated. The BioEstat 5.0 software was used, at significance level of 5% (P < 0.05).
RESULTS

Clinical evaluation

The dogs of the GCO and GAHT groups displayed average age of 4.8 ± 2.6 months and 3.4 ± 1.07 months and average weight of 3.96 ± 1.90 and 5.6 ± 5.0 kg, respectively. Males prevailed in both groups. Regarding the breeds, 50% of the dogs of the GCO were mongrel (ND), 20% Poodle, 20% Pinscher and 10% Cocker Spaniel while, in the GAHT, 60% were mongrel (ND), 20% Poodle and 20% Pinscher. Moreover, it was observed that of the dogs of the GCO and GAHT groups, respectively, 30% and 40% had incomplete immunization, and 70% and 60% were not vaccinated at all. As for worming, 7/10 of the GCO and 10/10 GAHT were not wormed.

The main clinical symptoms reported for the dogs of both groups were lethargy (20/20), anorexia (20/20), persistent vomiting (19/20) and foul-smelling diarrhea (20/20), varying in color from bloody (14/20) to yellow with traces of blood (6/20).

As for the vital parameters, no statistical differences between the experimental groups studied (Table 1) were observed. Regarding the remission of clinical signs, all parameters evaluated were significantly between the groups (Table 2). During the hospital stay, there was complete remission of clinical signs initially cited, but this was not homogeneous. Three dogs from the GCO needed to be treated for a longer period. Two of them presented endotoxemia and one died after 72 h and 70 h after initiating the treatment. In comparison, the animals of the GAHT exhibited the most effective clinical signs of recovery, evidenced by shortening by half the time required for recovery compared to GCO. The hospital discharge records show that from the animals of the GAHT, one was discharged 33 h and another two 46 h after admission.

In the GCO, three animals remained with signs of vomiting and diarrhea beyond the specified period of evaluation, thus requiring an additional 36 h ± 24 h of hospital treatment. Eight animals of the GCO required a total of 72 h ± 12.8 h to recover from the anorexia symptoms. These events were not observed outside of the evaluation period for the dogs of the GAHT.

With regard to treatment, six dogs from the GCO were required to undergo complementary therapy, which according to each clinical condition, consisted of Tranexamic acid (30 mg/kg), methylprednisolone (1 mg/kg), Sucralfate (30 mg/kg), Ampicillin (20 mg/kg), Ketoprofen (1 mg/kg), and metronidazole (15 mg/kg). Only one dog from the GAHT required complementary therapy, which consisted of adding Tranexamic acid (30 mg/kg) to the protocol. All animals underwent endoparasites control with Ivermectin 1% (0.2 mL/kg), and one animal from each group was treated for Babesiosis with imidocarb dipropionate (5 mg/kg).

<table>
<thead>
<tr>
<th>Variable</th>
<th>Group</th>
<th>M0</th>
<th>M12</th>
<th>M24</th>
<th>M36</th>
<th>M48</th>
<th>Normal range values*</th>
</tr>
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<tr>
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<td></td>
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<td></td>
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<tr>
<td>HR</td>
<td>GAHT</td>
<td>154 ± 44aA</td>
<td>149 ± 47aA</td>
<td>121 ± 31aA</td>
<td>137 ± 35aA</td>
<td>127 ± 31aA</td>
<td>60-160 bpm</td>
</tr>
<tr>
<td></td>
<td>GCO</td>
<td>160 ± 61aA</td>
<td>137 ± 18AA</td>
<td>124 ± 30aA</td>
<td>120 ± 40aA</td>
<td>114 ± 54aa</td>
<td></td>
</tr>
<tr>
<td>RR</td>
<td>GAHT</td>
<td>38 ± 22aA</td>
<td>48 ± 11aA</td>
<td>38 ± 10aA</td>
<td>40 ± 13 aA</td>
<td>38 ± 14 aA</td>
<td>18-36 mpm</td>
</tr>
<tr>
<td></td>
<td>GCO</td>
<td>34 ± 35aA</td>
<td>38 ± 58aA</td>
<td>30 ± 18aA</td>
<td>34 ± 33aa</td>
<td>36 ± 11aA</td>
<td></td>
</tr>
<tr>
<td>RT</td>
<td>GAHT</td>
<td>38.4 ± 0.4aA</td>
<td>38.2 ± 0.4aA</td>
<td>38.1 ± 0.5aA</td>
<td>38.4 ± 0.3aA</td>
<td>38.4 ± 0.4aA</td>
<td>37.5 to 39.5° C</td>
</tr>
<tr>
<td></td>
<td>GCO</td>
<td>38.5 ± 0.8aA</td>
<td>38.5 ± 1.6aA</td>
<td>38.3 ± 0.3aA</td>
<td>38.7 ± 0.7aA</td>
<td>38.2 ± 0.9aA</td>
<td></td>
</tr>
</tbody>
</table>

Lowercase letters correspond to the rows while the uppercase letters correspond to the columns. *Feitosa [8].
Table 2. Mean/median and standard deviation/interquartile range, in hours, of remission of clinical signs of dogs affected with the parvovirus, in the control group (GCO) and in the group submitted to autohemotherapy (GAHT).

<table>
<thead>
<tr>
<th>Symptom</th>
<th>Group</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>GCO</td>
<td>GAHT</td>
</tr>
<tr>
<td>Apathy</td>
<td>54 ± 42a*</td>
<td>24 ± 12b*</td>
</tr>
<tr>
<td>Anorexia</td>
<td>67 ± 15a**</td>
<td>36 ± 13b**</td>
</tr>
<tr>
<td>Vomiting</td>
<td>30 ± 51a*</td>
<td>12 ± 0b*</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>36 ± 21a*</td>
<td>12 ± 12b*</td>
</tr>
<tr>
<td>Dehydration</td>
<td>24 ± 21a*</td>
<td>12 ± 0b*</td>
</tr>
<tr>
<td>Pale mucous</td>
<td>54 ± 21a*</td>
<td>24 ± 12b*</td>
</tr>
<tr>
<td>Abdominal sensitivity</td>
<td>54 ± 28a**</td>
<td>19b ± 20**</td>
</tr>
<tr>
<td>Hospital discharge</td>
<td>70.8 ± 17.38a**</td>
<td>43.2 ± 10.11b**</td>
</tr>
<tr>
<td>Deaths (n/%)</td>
<td>1/10%</td>
<td>0</td>
</tr>
<tr>
<td>Endotoxemia(n/%)</td>
<td>2/20%</td>
<td>0</td>
</tr>
</tbody>
</table>

Lowercase letters correspond to the rows while uppercase letters correspond to the columns; Means followed by different letters are statistically different (P < 0.05) *Median; **Average.

**DISCUSSION**

There is a growing demand for more accurate and efficient therapeutic protocols to treat several infectious diseases that affect dogs, especially those with acute evolution, such as the parvovirus infection. Factors related to antimicrobial resistance, contraindications regarding age, cost, benefit, have hampered the use of many drugs, considerably decreasing treatment options, and leading to research for new alternatives. Thus, use of AHT in this study proved to be low-cost, accessible and easy to administer by the Veterinarian.

**Clinical evaluation**

In general, a higher prevalence of the disease was observed in young animals, with defined breed that had not been wormed and vaccinated. These facts show a greater exposure of these dogs to parvovirus infection, and minor, but worrying nonetheless, parasitosis. The low immunological capability, compromised or total lack of vaccination and poor sanitary conditions are the main factors that determine the appearance and installation of the disease [7,15]. From this study, it can be inferred that age and health conditions associated with deficient sanitation were major factors triggering and intensifying the infectious process.

The severity of infections caused by canine parvovirus (CPV), and consequently the time of remission of clinical signs vary independently of the protocol used, due to animal age [17]. It is suggested that the AHT protocol can be considered safe according to this research, since although younger and clinically relatively weaker, no animal of the GAHT died or presented symptoms of endotoxemia.

Additionally, the animals clinical debilitating initial picture with varied stool colors, especially the numerically more parasitized GAHT dogs, clearly showed a clinically more severe parasitic gastrointestinal infection. It is more debilitating and more difficult to treat associated infections [18,19]. Furthermore, as the CPV replicates only in dividing cells, any agent (parasitic, bacterial or viral) that causes destruction of the villi, stimulating mitosis in the intestinal tissue, can facilitate the reproduction of CPV, and, as a result, cause a more severe disease [3].

The vomiting and bloody diarrhea reported are closely associated with the erosive and inflammatory processes of the stomach and intestinal mucosa [14,15], where the vomiting originated in the secondary disturbances to the viral agent, resulting from the stimulation of peripheral receptors located on the TGI, activating the vomiting center located in the medulla [8], and the diarrheal process by lesions in the germinal epithelium of intestinal crypts [17]. These findings corroborate the considerations presented in the literature, which emphasize that vomiting and diarrhea are the first to be observed, causing the animals to become lethargic and depressed, as well as anorexia as the disease progresses [6].
With regard to the time of clinical remission, it is possible to correlate early improvement of vomiting and dehydration symptoms to both the effective action of Maropitant citrate [5] and the remission of inflammatory processes in the gastric mucosa promoted by the adopted support treatment. It can be inferred that such effects reduced energy consumption and abdominal discomfort caused by muscle contractions in the act of vomiting, as well as the hemodynamic improvement and correction of fluid and electrolyte imbalance promoted by dehydration [27].

Another important aspect to be highlighted concerns the recovery from diarrheal symptoms, which again was more effective in the GAHT. The AHT promotes the increase of leucocytes in abdominal organs [28]. It is, therefore, suggested that these cells may be present in greater amounts in the gut of this patients, providing a more intense and effective action, with earlier recovery of the pathological process, aiding, besides the recovery of diarrheal symptoms, the improvement in the general condition of the animal. Thus, it is believed that the treatment adopted in GAHT provided a dynamic retrieval of existing lesions in the germinal epithelium of the intestinal crypts, which are the cause of hemorrhagic diarrhea [14,17].

Finally, the remission of anorexia of the GAHT took approximately 50% of the time of the GCO. The anorexia remission is directly related to the recovery of the inflammatory process in the gastrointestinal tract (GIT). To this end, neuroendocrine mechanisms, such as the release of cytokines in the inflammatory processes, may be valuable for anorexia [22], further stressing that any disorder that reduces the brain stimulation, such as apathy, can also reduce food consumption.

The endotoxemia and the deaths observed, as well as complementary therapies added to the GCO protocol adopted, suggest that the adopted antibiotic therapy was not widely effective to treat the gastrointestinal symptoms and disease. This fact corroborates the results previously described for the treatment of dogs with parvovirus infection [11,20]. The absence of such symptoms in the GAHT strongly suggests the effectiveness of the autohemotherapy in controlling the infection, contributing to survival rates and clinical progress.

**CONCLUSION**

These results show that the autohemotherapy is clinically effective. They determine early recovery of patients suffering from parvovirus, without side effects or systemic organic impairment, revealing benefits as adjuvant alternative therapy to treat parvovirus successfully.

**SOURCES AND MANUFACTURERS**

1 SensPERT P®, Vencofarma Ltda., Brazil, PR, Brazil.
2 Eurofarma Laboratories Ltd., SP, Brazil.
3 Eurofarma Laboratories Ltd., SP, Brazil.
4 Cimetidine Hydrochloride ®, Teuto Brasileiro S/A, GO, Brazil.
5 Cerenia®, Pfizer Ltda. - Division of Animal Health, SP, Brazil.
6 Enrofloxacin®, Vencofarma Ltd. of Brazil, PR, Brazil.
7 Dipyrone D-500®, Fort Dodge, SP, Brazil.
8 Hertavita®, Hertape Calier, SP, Brazil.
9 Hemoblock®, Sigma Pharma Ltda, SP, Brazil.
10 Corti-dural®, König of Brazil Ltda, SP, Brazil.
11Sucrafilm®, Sigma Pharma, SP, Brazil.
12Ampicillin®, EMS S/A, SP, Brazil.
13 Ketoflex1%®, Vencofarma Ltd. of Brazil, PR, Brazil.
14Flagyl®, Sanofi Aventis Pharmaceutical Ltd., SP, Brazil.
15Baymec®, Bayer, SP, Brazil.
16Diazen®, MG, Brazil.

**Ethical approval.** This study was approved by the Ethics and Research Committee (COEP) of the Center for Health and Rural Technology, Universidade Federal de Campina Grande, Patos, PB, under protocol n° 28/2011.

**Declaration of interest.** The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

**REFERENCES**


